Filing Date: September 12, 2003

Title: METHOD AND KIT FOR IDENTIFYING VANCOMYCIN-RESISTANT ENTEROCOCCUS

In the Claims

Please amend the claims as follows:

- (Currently Amended) A method to detect vanA in a sample, comprising: 1.
- a) contacting a sample suspected of comprising amplified vanA nucleic acid with at least one vanA-specific oligonucleotide probe under high stringency hybridization conditions effective to form a hybrid between the vanA-specific oligonucleotide probe and vanA nucleic acid in the sample, wherein the vanA-specific oligonucleotide probe under high stringency conditions hybridizes to sequences which include sequences substantially corresponding to SEQ ID NO:3, the complement thereof, or a portion thereof and comprises sequences which include sequences substantially corresponding to nucleotides 870 to 896 (SEQ ID NO:3) of the vanA gene, the complement thereof, or a portion of the sequences substantially corresponding to nucleotides 870 to 896 or the complement thereof and wherein the amplified vanA nucleic acid has, sequences substantially corresponding to nucleotides 851 to 868 (SEQ ID NO:2) of the vanA gene, the complement thereof, or a portion of the sequences substantially corresponding to nucleotides 851 to 868 or the complement thereof, or and sequences substantially corresponding to nucleotides 898 to 917 (SEQ ID NO:4) of the vanA gene, the complement thereof, or a portion of the sequences substantially corresponding to nucleotides 898 to 917 or the complement thereof; and
- b) detecting or determining the presence or amount of hybrid formation, wherein hybrid formation is indicative of vanA nucleic acid in the sample.
- 2. (Withdrawn) A method to detect *vanB* in a sample, comprising:
- a) contacting a sample suspected of comprising amplified vanB nucleic acid with at least one vanB-specific oligonucleotide probe under high stringency hybridization conditions effective to form a hybrid between the vanB-specific oligonucleotide probe and vanB nucleic acid in the sample, wherein the vanB-specific oligonucleotide probe comprises sequences which include sequences substantially corresponding to nucleotides 387 to 404 of the vanB gene, the complement thereof, or a portion thereof, sequences substantially corresponding to nucleotides 406 to 423 of the vanB gene, the complement thereof, or a portion thereof, or sequences

substantially corresponding to nucleotides 426 to 446 of the vanB gene, the complement thereof, or a portion thereof; and

- b) detecting or determining the presence or amount of hybrid formation.
- (Withdrawn) A method to detect vanA in a sample, comprising: 3.
- a) contacting a biological sample suspected of comprising nucleic acid with at least one vanA-specific oligonucleotide primer under conditions effective to amplify vanA nucleic acid, wherein the vanA-specific oligonucleotide primer comprises sequences which include sequences substantially corresponding to nucleotides 870 to 896 of the vanA gene, the complement thereof, or a portion thereof, sequences substantially corresponding to nucleotides 851 to 868 of the vanA gene, the complement thereof, or a portion thereof, or sequences substantially corresponding to nucleotides 898 to 917 of the vanA gene, the complement thereof, or a portion thereof; and
 - b) detecting or determining the presence or amount of amplified nucleic acid.
- 4. (Withdrawn) A method to detect *vanB* in a sample, comprising:
- a) contacting a biological sample suspected of comprising nucleic acid with at least one vanB-specific oligonucleotide primer under conditions effective to amplify vanB nucleic acid, wherein the vanB-specific oligonucleotide primer comprises sequences which include sequences substantially corresponding to nucleotides 387 to 404 of the vanB gene, the complement thereof, or a portion thereof, sequences substantially corresponding to nucleotides 406 to 423 of the vanB gene, the complement thereof, or a portion thereof, or sequences substantially corresponding to nucleotides 426 to 446 of the vanB gene, the complement thereof, or a portion thereof; and
 - b) detecting or determining the presence or amount of amplified nucleic acid.
- 5. (Withdrawn) The method of claim 3 wherein one vanA-specific oligonucleotide primer comprises sequences corresponding to nucleotides 851 to 868 of the vanA gene or a portion thereof.

gene or a portion thereof.

6. (Withdrawn) The method of claim 3 wherein one *vanA*-specific oligonucleotide primer comprises sequences corresponding to the complement of nucleotides 898 to 919 of the *vanA*

- 7. (Withdrawn) The method of claim 3 wherein the presence or amount of amplified nucleic acid is detected or determined with an oligonucleotide probe comprising sequences corresponding to nucleotides 870 to 896 of the *vanA* gene, the complement thereof or a portion thereof.
- 8. (Currently Amended) The method of claim 1 wherein one the *vanA*-specific oligonucleotide probe comprises sequences corresponding to nucleotides 870 to 896 of the *vanA* gene, the complement thereof or the portion thereof is no more than 50 nucleotides in length and has at least 10 contiguous nucleotides of SEQ ID NO:3 or the complement thereof.
- 9. (Currently Amended) The method of claim 8 wherein the amplified nucleic acid is obtained by amplifying a biological sample comprising nucleic acid with at least one *vanA*-specific oligonucleotide primer comprising sequences corresponding to nucleotides 851 to 868 of the *vanA* gene SEQ ID NO:2 or the portion thereof, or sequences corresponding to the complement of nucleotides 898 to 917 of the *vanA* gene SEQ ID NO:4 or the portion thereof.
- 10. (Withdrawn) The method of claim 4 wherein one *vanB*-specific oligonucleotide primer comprises sequences corresponding to nucleotides 387 to 404 of the *vanB* gene or a portion thereof.
- 11. (Withdrawn) The method of claim 4 wherein one *vanB*-specific oligonucleotide primer comprises sequences corresponding to the complement of nucleotides 426 to 446 of the *vanB* gene or a portion thereof.
- 12. (Withdrawn) The method of claim 4 wherein the presence or amount of amplified nucleic acid is detected or determined with an oligonucleotide probe comprising sequences

corresponding to nucleotides 406 to 423 of the vanB gene, the complement thereof or a portion thereof.

- 13. (Withdrawn) The method of claim 2 wherein one vanB-specific oligonucleotide probe comprises sequences corresponding to nucleotides 406 to 423 of the vanB gene, the complement thereof or a portion thereof.
- (Withdrawn) The method of claim 13 wherein the amplified vanB nucleic acid is 14. obtained by amplifying a biological sample comprising nucleic acid with at least one vanBspecific oligonucleotide primer comprising sequences corresponding to nucleotides 387 to 404 of the vanB gene or a portion thereof, or sequences corresponding to the complement of nucleotides 426 to 446 of the *vanB* gene or a portion thereof.
- 15. (Previously Presented) The method of claim wherein the sample is a physiological sample.
- 16. (Original) The method of claim 15 wherein the sample is a peri-rectal sample.
- 17. (Previously Presented) The method of claim 1 or 8 further comprising contacting a corresponding sample with a probe which is not a *vanA*-specific probe.
- 18. (Previously Presented) The method of claim 1 or 8 further comprising contacting the sample with a probe which is not a *vanA*-specific probe.
- 19. (Original) The method of claim 17 or 18 further comprising comparing the presence or amount of hybrid formation with the vanA-specific oligonucleotide probe to the presence or amount of hybrid formation between the sample contacted with the non-vanA probe.
- 20. (Withdrawn) The method of claim 2, 12, or 13 further comprising contacting a corresponding sample with a probe which is not a *vanB*-specific probe.

21. (Withdrawn) The method of claim 2, 12, or 13 further comprising contacting the sample

with a probe which is not a *vanB*-specific probe.

22. (Withdrawn) The method of claim 20 or 21 further comprising comparing the presence

or amount of hybrid formation with the vanB probe to the presence or amount of hybrid

formation between the sample contacted with the non-vanB probe.

23. (Original) The method of claim 17 or 18 wherein the non-vanA probe is a vanB-specific

probe.

24. (Withdrawn) The method of claim 20 or 21 wherein the non-vanB probe is a vanA-

specific probe.

25. (Previously Presented) The method of claim 8, wherein the probe is labeled.

26. (Withdrawn) The method of claim 23 wherein the vanA-specific probe is labeled with a

different label than the vanB-specific probe.

27. (Withdrawn) The method of claim 24 wherein the vanB-specific probe is labeled with a

different label than the vanA-specific probe.

28. (Withdrawn) A method to detect vanA nucleic acid and vanB nucleic acid in a sample,

comprising:

a) contacting a sample suspected of comprising amplified vanA nucleic acid or amplified

vanB nucleic acid with at least one vanA-specific oligonucleotide probe and with at least one

vanB-specific oligonucleotide probe under high stringency hybridization conditions effective to

form a hybrid between the vanA-specific oligonucleotide probe and amplified vanA nucleic acid

and between the vanB-specific oligonucleotide probe and amplified vanB nucleic acid, wherein

the vanA-specific oligonucleotide probe comprises sequences which include sequences

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substantially corresponding to nucleotides 870 to 896 of the vanA gene, the complement thereof, or a portion thereof, sequences substantially corresponding to nucleotides 851 to 868 of the vanA gene, the complement thereof, or a portion thereof, or sequences substantially corresponding to nucleotides 898 to 917 of the vanA gene, the complement thereof, or a portion thereof, and wherein the vanB-specific oligonucleotide probe comprises sequences which include sequences substantially corresponding to nucleotides 387 to 404 of the vanB gene, the complement thereof, or a portion thereof, sequences substantially corresponding to nucleotides 406 to 423 of the vanB gene, the complement thereof, or a portion thereof, or sequences substantially corresponding to nucleotides 426 to 446 of the vanB gene, the complement thereof, or a portion thereof; and

- b) detecting or determining the presence or amount of hybrid formation.
- 29. (Withdrawn) A method to detect vanA nucleic acid and vanB nucleic acid in a sample, comprising:
- a) contacting a biological sample suspected of comprising vanA or vanB nucleic acid with at least one vanA-specific oligonucleotide primer under conditions effective to amplify vanA nucleic acid and with at least one vanB-specific oligonucleotide primer under conditions effective to amplify vanB nucleic acid, wherein the vanA-specific oligonucleotide primer comprises sequences which include sequences substantially corresponding to nucleotides 870 to 896 of the vanA gene, the complement thereof, or a portion thereof, sequences substantially corresponding to nucleotides 851 to 868 of the vanA gene, the complement thereof, or a portion thereof, or sequences substantially corresponding to nucleotides 898 to 917 of the vanA gene, the complement thereof, or a portion thereof, and wherein the vanB-specific oligonucleotides primer comprises sequences which include sequences substantially corresponding to nucleotides 387 to 404 of the vanB gene, the complement thereof, or a portion thereof, sequences substantially corresponding to nucleotides 406 to 423 of the vanB gene, the complement thereof, or a portion thereof, or sequences substantially corresponding to nucleotides 426 to 446 of the vanB gene, the complement thereof, or a portion thereof; and
 - b) detecting or determining the presence or amount of amplified nucleic acid.

- 30. (Withdrawn) The method of claim 29 wherein the presence or amount of amplified nucleic acid is detected with at least one *vanA*-specific oligonucleotide probe and at least one *vanB*-specific oligonucleotide probe.
- 31. (Withdrawn) The method of claim 28 or 30 wherein the at least one *vanA*-specific oligonucleotide probe and the at least one *vanB*-specific oligonucleotide probe have different labels.
- 32. (Withdrawn) An oligonucleotide composition comprising a first oligonucleotide comprising sequences substantially corresponding to nucleotides 870 to 896 of the *vanA* gene, the complement thereof, or a portion thereof, an oligonucleotide comprising sequences substantially corresponding to nucleotides 851 to 868 of the *vanA* gene the complement thereof, or a portion thereof, an oligonucleotide comprising sequences substantially corresponding to nucleotides 898 to 917 of the *vanA* gene, the complement thereof, or a portion thereof, or a combination thereof, wherein the oligonucleotide hybridizes under stringent hybridization conditions to *vanA* DNA.
- 33. (Withdrawn) An oligonucleotide composition comprising an oligonucleotide comprising sequences substantially corresponding to nucleotides 387 to 404 of the *vanB* gene, the complement thereof, or a portion thereof, an oligonucleotide comprising sequences substantially corresponding to nucleotides 406 to 423 of the *vanB* gene the complement thereof, or a portion thereof, an oligonucleotide comprising sequences substantially corresponding to nucleotides 426 to 446 of the *vanB* gene, the complement thereof, or a portion thereof, or a combination thereof, wherein the oligonucleotide hybridizes under stringent hybridization conditions to *vanB* DNA.
- 34. (Withdrawn) The oligonucleotide composition of claim 32 wherein at least one oligonucleotide has the length and sequence of any of SEQ ID NOs:2-4.
- 35. (Withdrawn) The oligonucleotide composition of claim 33 wherein at least one oligonucleotide has the length and sequence of any of SEQ ID NOs:6-9.

- 36. (Withdrawn) The oligonucleotide composition of claim 32 or 33 wherein the oligonucleotide is labeled.
- 37. (Withdrawn) A kit comprising an oligonucleotide specific for a *vanA* gene and/or a *vanB* gene in a test sample, comprising an oligonucleotide comprising sequences substantially corresponding to nucleotides 870 to 896 of the *vanA* gene, the complement thereof, or a portion thereof, or an oligonucleotide comprising sequences substantially corresponding to nucleotides 406 to 423 of the *vanB* gene, the complement thereof, or a portion thereof, wherein the oligonucleotide hybridizes under stringent hybridization conditions to *vanA* DNA or *vanB* DNA.
- 38. (Withdrawn) The kit of claim 37 further comprising at least one non-vanA or one non-vanB probe.
- 39. (Withdrawn) The kit of claim 37 further comprising an oligonucleotide comprising sequences substantially corresponding to nucleotides 387 to 404 of the *vanB* gene, the complement thereof, or a portion thereof, or an oligonucleotide comprising sequences substantially corresponding to nucleotides 426 to 446 of the *vanB* gene, the complement thereof, or a portion thereof.
- 40. (Withdrawn) The kit of claim 37 further comprising an oligonucleotide comprising sequences substantially corresponding to nucleotides 851 to 868 of the *vanA* gene, the complement thereof, or a portion thereof, or an oligonucleotide comprising sequences substantially corresponding to nucleotides 868 to 917 of the *vanA* gene, the complement thereof, or a portion thereof, or a combination thereof.
- 41. (Withdrawn) The kit of claim 37 wherein at least one oligonucleotide is labeled.
- 42. (Withdrawn) A kit comprising one or more oligonucleotides specific for a *vanA* gene in a test sample, comprising: an oligonucleotide comprising sequences substantially corresponding

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to nucleotides 851 to 868 of the vanA gene, the complement thereof, or a portion thereof, or an oligonucleotide comprising sequences substantially corresponding to nucleotides 898 to 917 of the vanA gene, the complement thereof, or a portion thereof, or a combination thereof.

43. (Withdrawn) A kit comprising one or more oligonucleotides specific for a vanB gene in a test sample, comprising: an oligonucleotide comprising sequences substantially corresponding to nucleotides 645 to 645 of the vanB gene, the complement thereof, or a portion thereof, or an oligonucleotide comprising sequences substantially corresponding to nucleotides 426 to 446 of the vanB gene, the complement thereof, or a portion thereof, or a combination thereof.